POSTER COMMUNICATIONS

A sensitive gas chromatographic technique for quantification of urinary tryptamine

G.B. BAKER, D.G. CALVERLEY, W.G. DEWHURST & I.L. MARTIN

Department of Psychiatry, University of Alberta, Edmonton, Canada and MRC Neuropharmacology Unit, The Medical School, Birmingham, UK

The involvement of tryptamine has been suggested in a variety of psychiatric disorders, particularly abnormal mood. Urinary tryptamine has been reported to be reduced in depressed patients (Rodnight, 1961; Coppen, Shaw, Malleson, Eccleston & Gundy, 1965) and to increase on recovery from depression (Coppen et al., 1965) and after treatment with phenelzine (Dewhurst, 1968). We have developed a simple gas chromatographic technique, using electron capture detection, for measurement of tryptamine in 1 ml of human urine. This technique is a modification of a procedure previously reported for measurement of rat brain phenylethylamine (Martin & Baker, 1977).

To 1 ml urine is added 5-methyltryptamine (100 ng) as internal standard, and the pH is adjusted to 7.8. After centrifugation, 100 µl phosphate buffer (0.2 M, pH 7.8) is added to the supernatant, which is then shaken for 1 min with 2 ml diethylhexylphosphoric acid in chloroform (2.5% v/v). Following brief centrifugation, the aqueous layer is aspirated off and the chloroform layer is removed to a clean test tube and shaken for 2 min with 2 ml HCl (0.5 M). A 1.5 ml aliquot of the HCl is removed, neutralized with solid sodium bicarbonate, acetylated using acetic anhydride and extracted with 3 ml ethyl acetate. The ethyl acetate is removed and taken to dryness under a stream of nitrogen. The residue is taken up in 25 µl ethyl acetate and reacted with pentafluoropropionic anhydride at 60°C for 30 minutes. The reaction mixture is allowed to cool and partitioned between 300 µl cyclohexane and 3 ml saturated sodium tatraborate. After centrifugation, the cyclohexane layer is retained and a 2 µl aliquot is used for gas chromatographic analysis (column: 5% OV-7 on Chromosorb 750; oven temp: 200°C; flow rate: 40 ml/min). Tryptamine and 5-methyltryptamine form β -carboline derivatives with retention times of 4.4 and 5.8 min respectively.

The procedure gives high consistent overall extractions, and the resulting derivatives have excellent chromatographic characteristics.

This technique has been used to determine the tryptamine levels in 24 h urine samples from 12 healthy volunteers (6 male and 6 female). The mean tryptamine excretion (\pm s.e. mean) was 79 (\pm 20) μ g/24 h which is consistent with control levels reported using other types of techniques (Sjoerdsma, Oates, Zaltzman & Udenfriend, 1959; Rodnight, 1961; Slingsby & Boulton, 1976).

The reported technique is rapid, inexpensive, requires only 1 ml of a urine sample and allows for simultaneous quantification of urinary tryptamine from a large number of subjects. It therefore offers distinct advantages over techniques previously available.

Financial support was provided by the Medical Research Council of Canada, the Alberta Mental Health Research Fund and the Special Services and Research Committee, University of Alberta Hospital. Dr R. Coutts performed the mass spectrometric analysis of the derivaties.

References

COPPEN, A., SHAW, D.M., MALLESON, A., ECCLESTON, E. & GUNDY, G. (1965). Tryptamine metabolism in depression. *Br. J. Psychiat.*, 111, 993-998.

DEWHURST, W.G. (1968). New theory of cerebral amine function and its clinical application. *Nature*, *Lond.* 218, 1130-1133.

MARTIN, I.L. & BAKER, G.B. (1977). A gas-liquid chromatographic method for the estimation of 2-phenylethylamine in rat brain tissue. *Biochem. Pharmac.*, 26, 1513-1516.

RODNIGHT, R. (1961). Body fluid indoles in mental illness. *Int. Rev. Neurobiol.* 3, 251-292.

SJOERDSMA, A., OATES, J.A., ZALTZMAN, P. & UDENFRIEND, S. (1959). Identification and assay of urinary tryptamine: application as an index of monoamine oxidase inhibition in man. J. Pharmac. exp. Ther., 126, 217-222.

SLINGSBY, J.M. & BOULTON, A.A. (1976). Separation and quantification of some urinary arylalkylamines. J. Chromatogr., 123, 51-56.